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Review

Antimicrobial activity of essential oils extracted from medicinal plants against the pathogenic microorganisms: A review

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Essential oils are complex volatile compounds, naturally synthesized by various parts of the plant during the secondary metabolism of plants. A wide range plants having the medicinal properties have been explored and used for the extraction of essential oils worldwide due to their antimicrobial properties against the bacterial, fungal and viral pathogens. The presence of a large number of alkaloids, phenols, terpenes derivatives compounds and other antimicrobial compounds makes the essential oils more précised in their mode action against the ample variety of pathogenic microorganisms. Thus, the essential oils could be used as better supplements or alternatives against the pathogenic microorganisms. The aim of this review article is to focus on the antimicrobial activities of essential oils secreted by medicinal plants and the mechanisms involved in the inhibition of these pathogenic microorganisms.

Key words: Antimicrobial property, bacterial cell wall, pathogenic microorganisms, volatile oils

INTRODUCTION

Medicinal plants have the ability to inhibit the growth of wide range of pathogenic microorganisms due to presence of essential oils. The antimicrobial impact of essential oils and its various components extracted from medicinal plants has been well documented (Hammer et al., 2002; Hood et al., 2003; Duschatzky et al., 2005). Essential oils have been extracted from complex mixture of volatile molecules produced by the secondary metabolism of medicinal plants. Hammer et al., (1999) reported that the essential oils extracted from medicinal plants contain approximately 20-60 components of quite different concentrations.

Essential oils are natural, volatile liquid, complex compounds characterized by a strong odor, rarely colored, soluble in lipid and organic solvents. It could be synthesized by all plant organs, i.e. buds, flowers, leaves, stems, twigs, seeds, fruits, roots, wood or bark, and are stored in secretary cells, cavities, canals, epidermic cells or glandular trichomes (Bozin et al., 2006). Essential oils generally have 2-3 major components at fairly high concentrations (20-70%) compared to other components present in trace amount. For example, Carvacrol (30%) and thymol (27%) are the main components of the *Origanum compactum* essential oil (Betts, 2001).

The major components include two groups of distinct bio-synthetically origin, which may determined the biological activity against the pathogenic microorganisms (Pichersky et al., 2006). The majority of essential oils are composed of terpenes and terpenoids and other aromatic and aliphatic constituents, all characterized by low molecular weight. Terpenes are the major group of plant natural products characterized by an extensive variety of structural types and the most valuable compounds (Degenhardt et al., 2009). The terpene compounds are hydrocarbons of general formula $(C_5H_8)n$ formed from isoprene units. These compounds could be acyclic,

monocyclic, bicyclic or tricyclic (Abed, 2007). On the basis of diversity in their chemical structure, they could be classified into several groups as monoterpenes (C_{10}), sesquiterpenes (C_{15}), and diterpenes (C_{20}). The majority of the components of essential oils are monoterpenes represent approximately 90% of the essential oils. These are generally volatile in nature with pleasant odor (Bakkali et al., 2008).

The chemical profile of essential oils varies in the number of molecules, stereochemical properties of molecules, and also depends on the type of extraction. The extraction products may vary in quality, quantity and in composition according to climate, soil composition, plant organ, age and vegetative cycle stage (Masotti et al., 2003; Angioni et al., 2006). Essential oils or some of their constituents are indeed effective against a large variety of organisms including bacteria and viruses (Duschatzky et al., 2005), fungi (Hammer et al., 2002) and protozoa (Monzote et al., 2006). The essential oils are known for their bactericidal, virucidal, fungicidal activity due to their medicinal properties against the wide range of pathogenic microorganisms. Use of synthetic chemicals for the control of pathogenic microorganisms is limited because of their carcinogenic effect, acute toxicity and environmental hazards. The use of essential oils to combat with the infectious microrganisms and to control epidemic multi-drug resistant microorganisms is the promising approach (Mulyaningsih et al., 2010).

Antimicrobial activity of essential oils

A variety of essential oils have been screened for their antimicrobial activity (Cantrell et al., 1998) (Figure 1) The antimicrobial activity of plant-derived essential oils is the basis of many applications, especially in food preservation, aromatherapy and medicine. Cowan (1999) reported that approximately 3,000 essential oils are currently known so far. Out of which 300 are of commercially important and widely used in the pharmaceutical, agronomic, food, sanitary, cosmetic and perfume industries (Hajhashemi et al., 2003; Perry et al., 2003).

Antibacterial actions of essential oils

Conner (1993) found that cinnamon, clove, pimento, thyme, oregano, and rosemary plants had strong inhibitory effect against several bacterial pathogens. It has been also reported that essential oils extracted from some medicinal plants had the antibacterial effects against all the five tested food borne pathogens due to presence of phenolic compounds such as carvacrol, eugenol and thymol (Kim et al., 1995). However, Ramos-Nino et al., (1996) found that benzoic acids, benzaldehydes and cinnamic acid were able to inhibit the growth of *Listeria monocytogenes*. Similarly, Ouattara et al.,(1997) observed the antibacterial activity of selected spices on the meat spoilage bacteria.

Arora and Kaur (1999) analyzed the antimicrobial activity of garlic, ginger, clove, black pepper and green chilli on the human pathogenic bacteria viz. *Bacillus sphaericus, Enterobacter aerogenes*, *E. coli*, *P. aeruginosa*, *S. aureus*, *S. epidermidis*, *S. typhi* and *Shiguella flexneri* and stated that amongst all the tested spices, aqueous garlic extracts was sensitive against all the bacterial pathogens. Similarly, effect of clove extracts on the production of verotoxin by enterohemorrhagic *Escherichia coli* O157:H7 was investigated by Sakagami et al., (2000) and it was evident from the study that the verotoxin production was inhibited by clove extract. However, Elgayyar et al., (2001) examined the effectiveness of cardamom, anise, basil, coriander, rosemary, parsley, dill and angelica essential oil for controlling the growth and survival of pathogenic and saprophytic microorganisms. The results of their study showed that essential oils extracted oregano, basil and coriander plants have inhibitory effect against *Pseudomonas aeruginosa, S. aureus* and *Yersinia enterocolitica*.

Sakandamis et al., (2002) observed the effect of oregano essential oils on the behavior of *Salmonella typhimurium* in sterile and naturally contaminated beef fillets stored under aerobic and modified atmospheres. They have concluded that the addition of oregano essential oils checked the reduction in initial population of the tested bacterial pathogens. However, Hood et al., (2003) reported that the bacterial growth may be inhibited by the ample application of essential oils or their use at high concentrations and their mode of action results in decline of the bacterial cells. Similarly, Sokovic et al., (2009) observed the antibacterial activity of essential oils extracted from thyme and mint leaves against the *Staphylococcus aureus, Salmomella typhimurium* and *Vibrio parahaemolyticus*. The result showed that all the plants have antibacterial activity against the tested pathogens but the effect of thyme leaves extract was more pronounced compared to other plants. Moreover, Shan et al., (2011) showed cinnamon, oregano, clove, pomegranate peel, and grape seed were found effective against *S. enterica* at room temperature, but the clove extracts possess highest antibacterial activity. The antibacterial activity of essential oils extracted from medicinal has been summarized in Table 1.

Antifungal actions of essential oils

The essential oils and their components have been used broadly against moulds. The essentials oils extracts from many plants such as basil, citrus, fennel, lemon grass, oregano, rosemary and thyme have shown their considerable antifungal activity against the wide range of fungal pathogens (Kivanc, 1991). Arora and Kaur (1999) observed the sensitivity essential oil of spices against some fungal pathogens and concluded that garlic and clove

Figure 1. Some important medicinal plants widely used for the extraction of essential oils. A) Cinnamon (*Cinnamomum zeylancium*); B) Thyme (*Thymus broussonetii*); C) Rosmery (*Rosmarinus officinalis*) D) Oregano (*Origanum vulgare*); E) Clove (*Syzygium aromaticum*); F) Worm wood (*Artemisa arbrescens*); G) Caraway (*Carum carvic*); H) Lemon grass (*Cymbopogon citratus*); I) Sage (*Salvia officinalis*).

extracts had the strong ability to inhibit the growth of *Candida acutus, C. albicans, C. apicola, C. catenulata. C. inconspicua, C. tropicalis, Rhodotorula rubra, Sacharomyces cerevisae* and *Trignopsis variabilis*. However, Delaquis and Mazza (1995) reported the antimicrobial effects of isothiocyanate isolated from onion and garlic plants and stated that isothiocyanates may inactivate the extracellular enzymes through the oxidative cleavage of disulphide bonds. Similarly, Grohs and Kunz (2000) observed that mixtures of ground spices were effective to inhibit the growth of *C. lipolytica*. According to the report of Ultee and Smid (2001) oregano and thyme essential oils are apparently amongst the best inhibitors of fungal pathogens because of the presence of the phenolic compounds such as carvacrol and thymol as main constituents which might disrupt the fungal cell membrane. Antifungal activity of essential oils and its derivatives has been studied on viable cells count, mycelia growth and mycotoxins producing ability of moulds by Juglal et al., (2002) and concluded that amongst all tested essential oils clove, cinnamon and oregano essential oils are effective against the *Aspergillus parasiticus* and *Fusarium moniliforme*. However, McMahon et al., (2007) indicated that the formation of reactive thiocyanate radicals having antimicrobial property. The fungal activity of essential oils extracted from medicinal plants has been summarized in Table 1.

Antiviral actions of essential oils

The antiviral activity of essential oils were tested against many enveloped RNA and DNA viruses, such as herpes simplex virus type 1 and type 2 (DNA viruses), dengue virus type 2 (RNA virus), and influenza virus (RNA virus). However, essential oils extracted from oregano and clove were also tested against non-enveloped RNA and DNA viruses, such as adenovirus type 3 (DNA virus), poliovirus (RNA virus), and coxsackievirus B1 (RNA virus) (Wagstaff et al., 1994).

Melissa officinalis essential oils could also inhibit the replication of HSV-2, due to the presence of citral and citronellal (Allahverdiyev et al., 2004). The ability of replication of HSV-1 could be suppressed by the *in-vitro*

Table 1. Effect of essential oils extracted from medicinal plants on the pathogenic microorganisms

Plant	Part used	Chemical compounds	Inhibited Microorganisms	References
Cymbopogon citrates *Allium sativum*	Fruit Bulb	Ethanolic compounds Ethanolic compounds Isothiocynate	*Enteriobacteriacae, S. aureus* *Enteriobacteriacae, Candida* spp.	Grohs and Kunz, 2000
Thymus vulgaris	Arial part	Thymol, Linalol, Carvacrol	*L. monocytogens, E. coli, S. typhimirium, S. aureus*	Lambert et al., 2001
Pimpinella anisum	Seed	Trans-anethole	*S. typhimirium, E. coli*	Elgayyar et al., 2001
Origanum vulgare	Arial part	Carvacrol, Thymol, γ-Terpinene	*L. monocytogens, E. coli,* Adeno virus, Polio virus	Ultee and Smid, 2001
Feoniculum vulgare	Seed	Trans-anethole	*Alternaria alternata, Fusarium oxysporium, Aspergillus flavus*	Hammer et al., 2002
Cinnamomum zelancium	Bark	Cinnamaldehyde	*Enterobacteriacae*	Hood et al., 2003
Amomum kerervanh	Seed	Ethanolic compounds	*Enteriobacteriacae*	Burt, 2004
Syzygium aromaticum *Zingiber officinale*	Flower bud Rhizomes	Eugenol, Eugenylacetate Ethanolic compounds	*Eneriobactericae, A. fumigatus, Candida* spp., Adeno virus, Polio virus *Enteriobacteriacae*	Lanciotti et al., 2004
Artemisa arborescens	Leaves	β-Triketone	*Herpes simplex* virus	Sinico et al., 2005
Rosmarinus officinalis	Flower	Benzaylacetate, Linalool, α-pipene	*E.coli, S. typhimurm, B. cerus, S. aureus*	Oussalah et al., 2006
Thymus vulgaris, Mentha piperita	Arial part	1,8- Cineole, Eugenel	*S. aureus, S. typhimirium Vibrio parahaemolyticus*	Sokovic et al., 2009
Salvia officinalis	Arial part	1,8-Cineole-caphure, α-pipene	*S. aureus, E. coli*	Mulyaningsih et al., 2010
Verbana officinalis	Arial part	Borneol, Geranoil	*S. aureus, E. coli, S. typhimirium, L. monocytogens*	Shan et al., 2011

incubation of various essential oils. It is obvious from the study of Sinico et al., (2005) that the Herpes simplex virus type 1 (HSV-1), which is the cause of common viral infections in humans, such as mucocutaneous herpes infections, herpetic keratitis, herpetic encephalitis, and neonatal herpes could be strongly suppressed by the activity of essential oils extracted from *Artemisia arborescens*. Thus, it has been concluded that the essential oils have been frequently used as antiviral agents against several viral diseases in human (Koch et al., 2008) and it has a potential to be used as alternative to synthetic antiviral drugs (Baqui et al., 2001; Primo et al., 2001). The antiviral activity of essential oils extracted from medicinal plants has been summarized in Table 1.

Mechanism of action of essential oils against pathogenic microorganisms

Effects on bacterial cell wall

Antimicrobial actions of essential oils lead to the leaking of cell membrane and increased the membrane permeability (Lambert et al., 2001; Oussalah et al., 2006). The permeabilization of the cell membranes is directly

associated with loss of ions and reduction in membrane potential, collapse of the proton pump and depletion of the ATP pool (Di Pasqua et al., 2006; Turina et al., 2006). The disturbed cell structure may affect others cellular structures in a cascade type of action (Carson et al., 2002). Essential oils pass through the cell wall and cytoplasmic membrane may disrupt the structural arrangement of different polysaccharides, fatty acids and phospholipids layers (Burt, 2004; Longbottom et al., 2004). It may also coagulate in the cytoplasm and damage lipids and proteins layers (Burt, 2004)

Cytotoxic effects of essential oils were analyzed *in-vitro* experiments against most of pathogenic gram positive and gram negative bacteria not only confined to human or animal pathogens/parasites but also found effective in for the preservation of agricultural/marine products (Arnal-Schnebelen et al., 2004). The antimicrobial effect of essential oil components such as thymol, menthol and linalyl acetate might be due to a perturbation of the lipid fractions of bacterial plasma-membranes, which might be affected the membrane permeability and leakage of intracellular materials (Trombetta et al., 2005).

The other action of essential oils on the cell membrane is the inhibition of toxin secretion. Ultee and Smid (2001) reported that the exposure of *B. cereus* to carvacrol resulted on inhibition of diarrheal toxin production and use of oregano completely abolish the enterotoxin production of *S. aureus.* However, Ultee et al., (2000) reported that the secretion of toxins may be prevented by modifications in the bacterial membrane due to the attachment of the essential oil which might control the trans-membrane transport process across the plasma membrane and limit the release of toxins to the external environment (de Souza et al., 2010).

The disruption of the cell membrane by essential oils may help in various vital processes such as energy conversion processes, nutrient processing, synthesis of structural macromolecules, and secretion of many growth regulators (Oussalah et al., 2006). Moreover, Turina et al., (2006) emphasized that effect of specific ions due on plasma membrane has strong effect on the protons motive force, intracellular ATP content and overall activity of microbial cells such as turgor pressure, solutes transport and metabolism regulation process.

Effects on fungal cell wall

The essential oils have the ability to penetrate and disrupt the fungal cell wall and cytoplasmic membranes, permeablise them and finally damage mitochondrial membranes. The changes in electron flow through the electron transport system inside the mitochondria damage the lipids, proteins and nucleic acid contents (Arnal-Schnebelen et al., 2004) of the fungal cells. The essential oils could also hassle the depolarization of the mitochondrial membranes and decreasing the membrane potential, affect Ca^{2+} and other ion channels, reduce the pH and also affect the proton pump and ATP pool. The change the fluidity of membranes resulted into the leakage of radicals, cytochrome C, calcium ions and proteins. Thus, permeabilization of outer and inner mitochondrial membranes leads to cell death by apoptosis and necrosis (Yoon et al., 2000).

Conclusion

Most of the medicinal plants possess antimicrobial activity due to presence the essential oil. The nature, structural composition, and the functional groups present in the essential oils play an important role in determining the antimicrobial activity. Essential oils contain a variety of volatile molecules such as terpenes and terpenoids, phenol-derived aromatic and aliphatic compounds, which might have bactericidal, virucidal, and fungicidal consequences. Essential oils affect directly the cell membrane of the pathogenic microorganism by causing an increase in permeability and leakage of vital intracellular constituents, and finally disrupt the cell respiration and microbial enzyme system. Moreover, they also exhibited the cytotoxic effects on living cells due to their type and concentration. Therefore, it has been suggested that the essential oils extracts from the medicinal plants might be used as alternative antimicrobial natural substances and also play a great role in the discovery of new drugs.

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